analysis, add 10 milliliters of 0.9 percent saline solution and 1 milliliter of 0.1 percent bicarbonate solution. Add phosphate buffer, 0.001*M*, and shake until dissolved. Sonicate, if necessary, but for no longer than 1 minute. Dilute to volume with phosphate buffer, 0.001*M*, to obtain a solution containing approximately 500 micrograms of imipenem per milliliter. Mix well and inject immediately.

(*b*) *Sample solution.* Dissolve an accurately weighed portion (approximately 25 milligrams) of the sample with 10 milliliters of 0.9 percent saline solution and 1 milliliter of 0.1 percent bicarbonate solution in a 50-milliliter volumetric flask. Dilute the sample solution to volume with phosphate buffer, 0.001*M*, to obtain a solution containing 500 micrograms of imipenem per milliliter (estimated).

(iii) *System suitability requirements*—

(*a*) *Tailing factor.* The tailing factor (*T*) is satisfactory if it is not more than 1.5 at 10 percent of peak height in lieu of 5 percent of peak height.

(*b*) *Efficiency of the column.* The efficiency of the column (*n*) is satisfactory if it is greater than 600 theoretical plates for a 30-centimeter column.

(*c*) *Resolution.* The resolution (*R*) between the peaks for thienamycin and imipenem is satisfactory if it is not less than 2.0.

(*d*) *Coefficient of variation (relative standard deviation).* The coefficient of variation ($S_R$inpercent) of 5 replicate injections is satisfactory if it is not more than 2.0 percent.

If the system suitability requirements have been met, then proceed as described in §436.216(b) of this chapter. Alternate chromatographic conditions are acceptable provided reproducibility and resolution are comparable to the system. However, the sample preparation described in paragraph (b)(1)(ii)(*b*) of this section should not be changed.

(iv) *Calculations.* Calculate the micrograms of imipenem per milligram of sample as follows:

$$\text{Micrograms of imipenem per milligram} = \frac{A_u \times P_s \times 100}{A_s \times C_u \times (100 - L)}$$

where:

A_u=Area of the imipenem peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_s=Area of the imipenem peak in the chromatogram of the imipenem working standard;

P_s=Anhydrous imipenem activity in the imipenem working standard solution in micrograms per milliliter;

C_u=Milligrams of sample per milliliter of sample solution; and

L=Percent loss on drying of the sample.

(2) *Sterility.* Proceed as directed in §436.20 of this chapter, using the method described in paragraph (e)(1) of that section.

(3) *Pyrogens.* Proceed as directed in §436.32(a) of this chapter, using a solution containing 5.0 milligrams of imipenem per milliliter, except inject 10 milliliters per kilogram of rabbit weight.

(4) *Loss on drying.* Proceed as directed in §436.200(i) of this chapter.

(5) *Specific rotation.* Dilute an accurately weighed sample with sufficient pH 7.0 phosphate buffer to give a concentration of approximately 5.0 milligrams of imipenem per milliliter. Proceed as directed in §436.210 of this chapter, using a 1.0-decimeter polarimeter tube. To prepare the pH 7.0 phosphate buffer, transfer 5 grams of monobasic potassium phosphate and 11 grams of dibasic potassium phosphate to a 1.0-liter volumetric flask. Dissolve and dilute to volume with distilled water.

(6) *Identity.* Proceed as directed in §436.211 of this chapter, using the sample preparation described in paragraph (b)(2) of that section.

(7) *Crystallinity.* Proceed as directed in §436.203(a) of this chapter.

[51 FR 11573, Apr. 4, 1986; 51 FR 16517, May 5, 1986, as amended at 55 FR 11582, Mar. 29, 1990; 59 FR 8133, Feb. 18, 1994]

Subpart B—[Reserved]

Subpart C—Injectable Dosage Forms

§441.220 Imipenem monohydrate-cilastatin sodium injectable dosage forms.

§441.220a Sterile imipenem monohydrate-cilastatin sodium.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity.* Imipenem monohydrate-

cilastatin sodium is a dry mixture of imipenem monohydrate and cilastatin sodium packaged for dispensing. Its potency is satisfactory if it contains not less than 400 micrograms of imipenem and not less than 400 micrograms of cilastatin per milligram. Its imipenem content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of imipenem that it is represented to contain. Its cilastatin content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of cilastatin that it is represented to contain. It is sterile. It is nonpyrogenic. Its loss on drying is not more than 3.5 percent. When reconstituted as directed in the labeling, its pH is not less than 6.0 and not more than 7.5. The imipenem monohydrate used conforms to the standards prescribed by § 441.20a(a)(1).

(2) *Labeling.* It shall be labeled in accordance with the requirements of § 432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on:

(A) The imipenem monohydrate used in making the batch for potency, sterility, pyrogens, loss on drying, specific rotation, identity, and crystallinity.

(B) The batch for imipenem potency, cilastatin potency, imipenem content, cilastatin content, sterility, pyrogens, loss on drying, and pH.

(ii) Samples, if required by the Director, Center for Drug Evaluation and Research:

(A) The imipenem monohydrate used in making the batch: 10 packages, each containing approximately 500 milligrams.

(B) The batch:

(*1*) For all tests except sterility: A minimum of 20 immediate containers.

(*2*) For sterility testing: 20 immediate containers, collected at regular intervals throughout each filling operation.

(b) *Tests and methods of assay*—(1) *Imipenem and cilastatin potency and content.* Determine the potency of the sample in micrograms per milligram of both imipenem and cilastatin and the milligrams of both imipenem and cilastatin per container. Proceed as directed in § 441.20a(b)(1), preparing the cilastatin reference standard solution, the sample solution and calculating the imipenem and cilastatin potency and content as follows:

(i) *Cilastatin reference standard.* Accurately weigh approximately 25 milligrams of the cilastatin reference standard into a 50-milliliter volumetric flask. Immediately prior to analysis, add 10 milliliters of a 0.9 percent saline solution and 1.0 milliliter of a 0.1 percent bicarbonate solution. Add phosphate buffer, 0.001*M*, and shake until dissolved. Sonicate, if necessary, but no longer than 1 minute. Dilute to volume with phosphate buffer, 0.001*M*, to obtain a solution containing approximately 500 micrograms of cilastatin per milliliter. Mix well and inject immediately.

(ii) *Preparation of sample solutions*—(A) *Imipenem and cilastatin potency (micrograms of imipenem and cilastatin per milligram).* Remove the metal seal from each of 10 containers and determine the gross weight in grams. Dissolve and wash out the entire contents of each container with a 0.9 percent saline solution into an appropriate size volumetric flask to give a concentration of 5 milligrams per milliliter each of imipenem and cilastatin. Further dilute with phosphate buffer, 0.001*M*, to obtain a solution containing 500 micrograms each of imipenem and cilastatin per milliliter (estimated). Wash each stopper and container with small quantities of acetone or methanol three times being careful not to wet the container labeling. Allow the containers to air dry about 3 hours or to constant weight. Weigh each container and stopper to determine tare weight in grams.

(B) *Imipenem and cilastatin content (milligrams of imipenem and cilastatin per container).* Reconstitute the sample as directed in the labeling, except use a 0.9 percent saline solution as the reconstituting fluid. Then, using a suitable hypodermic needle and syringe, remove all of the withdrawable contents if it is represented as a single-dose container; or, if the labeling specifies the amount of potency in a given volume of the resultant preparation, remove an accurately measured representative portion from each container. Accurately dilute

the solution thus obtained in a suitable volumetric flask with sufficient 0.9 percent saline solution to obtain a stock solution containing about 2,500 micrograms of imipenem and 2,500 micrograms of cilastatin per milliliter. Transfer a 10-milliliter aliquot of this solution to a 50-milliliter volumetric flask and dilute to volume with phosphate buffer, 0.001*M*, to obtain a solution containing 500 micrograms of imipenem and 500 micrograms of cilastatin per milliliter (estimated).

(iii) *Calculations*—(A) *Imipenem and cilastatin potency.* Calculate the micrograms of imipenem and cilastatin per milligram as follows:

$$\text{Milligrams of imipenem or cilastatin per milligram} = \frac{A_U \times P_S \times d}{A_S \times 1{,}000 \times W_S}$$

where:

A_U=Area of the imipenem or cilastatin peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_S=Area of the imipenem or cilastatin peak in the chromatogram of the imipenem or cilastatin working standard;

P_S=Anhydrous imipenem or cilastatin activity in the respective working standard solution in micrograms per milliliter;

d=Dilution factor of the 10 samples; and

W_S=Net contents of 10 containers in grams (gross weight of 10 containers in grams—tare weight of 10 containers in grams).

(B) *Imipenem and cilastatin content.* Calculate the imipenem or cilastatin content of the container as follows:

$$\text{Milligrams of imipenem or cilastatin per milligram} = \frac{A_U \times P_S \times d}{A_S \times 1{,}000}$$

where:

A_U=Area of the imipenem or cilastatin peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_S=Area of the imipenem or cilastatin peak in the chromatogram of the imipenem or cilastatin working standard;

P_S=Anhydrous imipenem or cilastatin activity in the imipenem or cilastatin working standard solution in micrograms per milliliter; and

d=Dilution factor of the sample.

(2) *Sterility.* Proceed as directed in §436.20 of this chapter, using the method described in §436.20(e)(1).

(3) *Pyrogens.* Proceed as directed in §436.32(a) of this chapter, using a solution containing 5.0 milligrams of imipenem per milliliter except inject 10 milliliters per kilogram of rabbit weight.

(4) *Loss on drying.* Proceed as directed in §436.200(a) of this chapter.

(5) *pH.* Proceed as directed in §436.202 of this chapter.

[58 FR 26670, May 4, 1993]

§441.220b Imipenem monohydrate-cilastatin sodium for injection.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity.* Imipenem monohydrate-cilastatin sodium is a dry mixture of imipenem monohydrate, cilastatin sodium, and sodium bicarbonate packaged for dispensing. Its potency is satisfactory if it contains not less than 400 micrograms of imipenem and not less than 400 micrograms of cilastatin per milligram. Its imipenem content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of imipenem that it is represented to contain. Its cilastatin content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of cilastatin that it is represented to contain. It is sterile. It is nonpyrogenic. Its loss on drying is not more than 3.5 percent. When reconstituted as directed in the labeling, its pH is not less than 6.5 and not more than 8.5. The imipenem monohydrate used conforms to the standards prescribed by §441.20a(a)(1).

(2) *Labeling.* It shall be labeled in accordance with the requirements of §432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of §431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on:

(*a*) The imipenem monohydrate used in making the batch for potency, sterility, pyrogens, loss on drying, specific rotation, identity, and crystallinity.

(*b*) The batch for imipenem potency, cilastatin potency, imipenem content, cilastatin content, sterility, pyrogens, loss on drying, and pH.